ELSEVIER

Applied Soil Ecology 5 (1996) 21–27

Applied Soil Ecology

# Microbial diversity in the rhizosphere of corn grown under conventional and low-input systems

Jeffrey S. Buyer *, Donald D. Kaufman

*USDA, ARS, SMSL, Building 318 BARC-East, Beltsville, MD 20705-2350, USA*

Accepted 12 August 1996

**Abstract**

Microbial processes within the rhizosphere of crop plants are crucial to agriculture. The relation of soil microbial community diversity to cropping system, yield, and soil quality are unclear at present. The Farming Systems Trial at the Rodale Institute Research Center, Kutztown, PA, is a 15-year study in which a conventional corn–soybean rotation has been compared with low-input systems (i.e. animal manure or legumes as nitrogen sources). The effects of the cropping systems on diversity of fast-growing, aerobic, culturable bacteria and fungi were conducted in 1994 by taking 84 cores in between corn plants in June, July, and August. Rhizosphere bacteria and fungi were extracted, plated, and counted. Approximately 6000 bacteria were identified by fatty acid methyl ester analysis, while 18000 fungi were identified by microscopic examination of spores. Microbial diversity and evenness were calculated using several different methods. Total counts, diversity, and evenness were not significantly different for the three cropping systems. These results suggest that conventional agricultural practices may maintain high indices of microbial diversity in the rhizosphere. The functional significance of this needs to be investigated.

*Keywords:* Rhizosphere; Bacteria; Fungi; Biodiversity

## 1. Introduction

Alternative agricultural practices, including crop rotations, recycling of crop residues, increased use of cover crops and green manures, reduced chemical input, and integrated pest management, contribute to high soil organic matter levels and improve soil quality (Parr et al., 1992). Organic matter decomposition and nutrient cycling may be more important in sustainable than conventional agriculture, where chemical inputs dominate the system. Therefore, soil microorganisms are believed to play a crucial role in low-input sustainable agriculture (Parr et al., 1992).

Biological diversity is often used as an index of the health of an ecosystem, since environmental degradation often leads to lower diversity (Magurran, 1988, pp. 101–114). Soil microbiological diversity has been used as an index for soil quality, particularly in predicting the success of reclaiming severely degraded soils (Visser and Parkinson, 1992). Agricultural management practices might well have an impact on microbial diversity, which could in turn impact soil health, crop health and yield, and sustainability. According to one hypothesis a more complex soil microbial community will result in greater yield

* Corresponding author: Tel. 301-504-8436; Fax. 301-504-8370; Email. jbuyer@asrr.arsusda.gov.


*PII* S0929-1393(96)00127-8

stability (Cook and Baker, 1983). However, there is little direct evidence to support these concepts, largely because of the great difficulty in studying community structure and diversity of soil microorganisms (Lee, 1994; O'Donnell et al., 1994).

In this study we examine the population and diversity of bacteria and fungi isolated from the rhizosphere of corn (*Zea mays*) grown under both conventional and alternative agricultural systems.

## 2. Materials and methods

The Farming Systems Trial at the Rodale Institute Research Center in Kutztown, Pennsylvania, is a 15-year-old comparison of three cropping systems. The conventional treatment consists of a corn–soybean rotation using chemical fertilizer. The animal manure system produces red clover, alfalfa, oats, winter wheat, corn, and soybeans, using animal manure as the nitrogen source. The legume system relies on legume cover crops to supply nitrogen while producing corn, soybeans, oats, winter wheat, and spring barley. There are eight replicates for each treatment. Each replicate plot is approximately 91 m × 18 m, divided into 6.1 m × 6.1 m sampling plots.

Samples were taken in late June, late July, and late August of 1994. Two replicate plots (replicates 4 and 5) were sampled for each treatment. Seven adjacent sampling plots were used for each replicate plot. In each sampling plot, two cores (3 in. diameter, 6 in. deep) were taken in between two adjacent corn plants, and combined. Each core contained roots from the two adjacent plants so the combined samples included roots from four plants. Roots and adhering soil were separated from bulk soil, bagged, and placed in an insulated cooler. Each sample was transported to the laboratory within two hours and immediately processed. Bulk soil samples from the cores were bagged and stored at −20°C for fatty acid analysis (to be reported separately).

The roots and adhering soil were cut up and mixed. A subsample weighing approximately 10 g was transferred to a sterile tared 250 ml Erlenmeyer flask equipped with a sterile foam plug and weighed. The subsample was diluted 1:10 weight:volume with sterile saline buffered to pH 6.8 with 0.1 M 2-[(2-amino-2-oxoethyl)-amino]ethanesulfonic acid (ACES) and shaken for 10 min at 22°C. Another subsample was oven-dried and used to calculate percent moisture.

For enumeration and identification of bacteria the suspension was serially diluted with sterile buffer and dilution-plated using a Spiral Plater (Spiral Systems, Bethesda, MD). The isolation plates were prepared using Rhizosphere Isolation Medium (Buyer, 1995). Plates were incubated for 3 days at 22°C and counted using a Model 500A Bacterial Colony Counter with CASBA II software (Spiral Systems Instruments, Bethesda, MD). Forty-eight single colonies from each sample were transferred with sterile toothpicks to 96-well microplates containing 80 μl tryptic soy broth per well. The microplates were incubated at 28°C for a few hours, until most wells were visibly turbid, and then 20 μl of 50% glycerol were added to each well. The plates were stored at −80°C.

The plates were later thawed out and the contents of each well spread on a trypticase soy broth agar plate and incubated at 28°C. After overnight growth, single colonies were quadrant streaked on trypticase soy broth agar and incubated for 24 h at 28°C. Bacteria were identified by fatty acid methyl ester analysis using the MIDI system (Microbial ID, Inc., Newark, DE). Following identification, the isolates were suspended in trypticase soy broth containing 10% glycerol and stored at −80°C. Bacteria were divided into taxa by cluster analysis with dendrograms. Dendrograms were constructed with the single-link method using Dendrogram II (Microbial ID, Inc.). Isolates linked at an Euclidean distance of eight or less were considered to belong to the same taxon.

For enumeration and identification of fungi, serial dilutions of the rhizosphere suspension were plated on OAES agar (Kaufman et al., 1963) using a Spiral Plater. Plates were incubated at 24°C for four days. After incubation, each colony was numbered on the bottom of the plate. All fungi were identified as soon as adequate sporulation occurred. Identification was made to genus, with the exception of the aspergilli and penicilli, which were identified to groups and series according to Thom and Raper (1945) and Raper and Thom (1949). The gliocladia were identified to species as described by Gilman (1957). Where

Table 1
Bacterial and fungal population counts. Seven samples from each of two replicate plots were averaged. Values are mean ± standard deviation. Values within a column followed by different letters indicate significant differences ($P < 0.05$)

| Month | Treatment [a] | Bacterial count (log cfu $g^{-1}$ dry wt.) | Fungal count (log cfu $g^{-1}$ dry wt.) |
|---|---|---|---|
| Jun. | C | 6.84 ± 0.39bc | 4.72 ± 0.31c |
| | L | 6.68 ± 0.30c | 4.46 ± 0.41d |
| | A | 6.92 ± 0.25b | 4.82 ± 0.17d |
| Jul. | C | 7.44 ± 0.23a | 5.14 ± 0.25b |
| | L | 7.43 ± 0.19a | 5.28 ± 0.38ab |
| | A | 7.45 ± 0.21a | 5.37 ± 0.28a |
| Aug. | C | 6.70 ± 0.24c | 5.26 ± 0.20ab |
| | L | 6.86 ± 0.14bc | 5.30 ± 0.14ab |
| | A | 6.90 ± 0.12b | 5.28 ± 0.17ab |

[a] C, conventional; L, legume rotation; A, animal manure.

possible, identification was made on the original dilution plate but it was occasionally necessary to transfer a colony to another medium. In this case fresh potato dextrose agar was used. Several 'keys' were used in identification (Gilman, 1957).

Two different methods were used to calculate biodiversity and evenness (Pielou, 1969; Magurran, 1988). Shannon's index of diversity, $H'$, and index of evenness, $J'$, were calculated as

$$H' = -\sum_{i=1}^{S} \frac{n_i}{N} \ln \frac{n_i}{N}$$

$$J' = \frac{H'}{\ln S}$$

where $n_i$ represents the number of isolates in the $i$th taxon, $N$ is the total number of isolates, and $S$ is the total number of taxa. The McIntosh index of diversity

$$D = \frac{N - \sqrt{\sum_{i=1}^{S} n_i^2}}{N - \sqrt{N}}$$

and the McIntosh index of evenness

$$E = \frac{N\sqrt{N_i^2}}{N - \frac{N}{\sqrt{S}}}$$

were also used.

The taxonomic data for all seven samples within each replicate were combined to produce a biodiversity index for the replicate as a whole. For bacteria this was accomplished by constructing dendrograms for each replicate plot containing seven sampling plots. Each dendrogram had approximately 320 isolates. To make the data more manageable, only clusters consisting of two or more isolates were used for biodiversity calculations and for relative abundance of taxa.

Clusters were compared between dendrograms, first by comparing the names assigned to the isolates within the clusters by the MIDI system, and then by constructing new dendrograms containing isolates from each replicate plot believed to belong to the same taxa. This allowed us to classify isolates from each replicate plot into the same taxonomic groups.

Table 2
Diversity indices for bacteria calculated as the mean of two replicates. Values are mean ± standard deviation. Values within a column followed by different letters indicate significant differences ($P < 0.05$)

| Month | Treatment [a] | Diversity ($H'$) | Evenness ($J'$) | Diversity ($D$) | Evenness ($E$) |
|---|---|---|---|---|---|
| Jun. | C | 1.87 ± 0.04bcd | 0.64 ± 0.01e | 0.55 ± 0.02bcd | 0.68 ± 0.03bc |
| | L | 2.18 ± 0.26ab | 0.77 ± 0.05abc | 0.64 ± 0.05ab | 0.80 ± 0.04ab |
| | A | 2.02 ± 0.11abc | 0.75 ± 0.01abcd | 0.61 ± 0.02ab | 0.77 ± 0.00ab |
| Jul. | C | 2.11 ± 0.03abc | 0.77 ± 0.03abc | 0.62 ± 0.02ab | 0.78 ± 0.04ab |
| | L | 2.35 ± 0.26a | 0.79 ± 0.06a | 0.68 ± 0.06a | 0.83 ± 0.06a |
| | A | 2.26 ± 0.04a | 0.82 ± 0.07ab | 0.67 ± 0.02a | 0.84 ± 0.06a |
| Aug. | C | 1.78 ± 0.11cd | 0.69 ± 0.03cde | 0.56 ± 0.03bc | 0.72 ± 0.03abc |
| | L | 1.42 ± 0.14e | 0.66 ± 0.04de | 0.45 ± 0.08d | 0.65 ± 0.10c |
| | A | 1.54 ± 0.10de | 0.70 ± 0.04bcde | 0.50 ± 0.04cd | 0.70 ± 0.05bc |

[a] C, conventional; L, legume rotation; A, animal manure.

Table 3
Diversity indices for fungi calculated as the mean of two replicates. Values are mean ± standard deviation. Values within a column followed by different letters indicate significant differences ($P < 0.05$)

| Month | Treatment [a] | Diversity ($H'$) | Evenness ($J'$) | Diversity ($D$) | Evenness ($E$) |
|---|---|---|---|---|---|
| Jun. | C | 1.97 ± 0.03bcd | 0.76 ± 0.00ab | 0.59 ± 0.00abc | 0.74 ± 0.00ab |
| | L | 1.85 ± 0.08cde | 0.70 ± 0.04abc | 0.56 ± 0.01abc | 0.71 ± 0.01ab |
| | A | 1.99 ± 0.18bc | 0.77 ± 0.05a | 0.62 ± 0.05ab | 0.80 ± 0.06a |
| Jul. | C | 2.44 ± 0.16ab | 0.86 ± 0.03a | 0.71 ± 0.03a | 0.88 ± 0.02a |
| | L | 1.49 ± 0.54de | 0.51 ± 0.22d | 0.37 ± 0.20d | 0.46 ± 0.26c |
| | A | 2.51 ± 0.01a | 0.84 ± 0.02a | 0.72 ± 0.00a | 0.88 ± 0.00a |
| Aug. | C | 1.73 ± 0.15cde | 0.58 ± 0.05bcd | 0.45 ± 0.05bcd | 0.56 ± 0.05bc |
| | L | 1.67 ± 0.03cde | 0.56 ± 0.05cd | 0.43 ± 0.01cd | 0.54 ± 0.03bc |
| | A | 1.47 ± 0.07e | 0.49 ± 0.02d | 0.34 ± 0.02d | 0.42 ± 0.03c |

[a] C, conventional; L, legume rotation; A, animal manure.

Contradictory results were sometimes observed. For example, isolates A and B might be assigned to the same taxon, isolates A and C to the same taxon, but B and C to different taxa. In these cases we generally used broader definitions of taxa to produce fewer taxa rather than more, and thus in the example, A, B, and C would all be considered to belong to the same taxon.

Taxonomic abundance data were used to compare plants by principal coordinate analysis using the program NTSYS-PC (Exeter Software, Setauket, NY).

## 3. Results

Bacterial counts were not significantly different among the three treatments (Table 1). There was a significant time effect, with July having the highest populations. Fungal counts showed a significant increase from June to July (Table 1) in all three treatments. However, fungal counts did not decrease in August as did the bacterial counts.

Diversity and evenness were lowest for the bacteria in August (Table 2). The conventional treatment showed less seasonal effect than the legume and animal manure treatments. The fungi (Table 3) showed a decrease in diversity and evenness from July to August, but in this case the legume treatment showed the least seasonal effect.

A total of 92 bacterial taxa with two or more isolates were identified. Major bacterial taxa are shown in Table 4. The most dominant bacterial taxon was *Pseudomonas* (primarily *Pseudomonas chlororaphis*), followed by *Arthrobacter*. The *Arthrobacter* cluster also contained some *Bacillus* species. A cluster consisting only of *Bacillus megaterium* subgroup A was consistently separated from other *Bacil-*

Table 4
Fractional abundance of major bacterial taxa. Values are mean ± standard deviation. Values within a column followed by different letters indicate significant differences ($P < 0.05$)

| Month | Treatment [a] | *Agrobacteria* | *Arthrobacter* | *Bacillus megaterium* A | *Xanthomonas* | *Acinetobacter* | *Pseudomonas* | Enterics | *E. coli* |
|---|---|---|---|---|---|---|---|---|---|
| Jun. | C | 0.00 ± 0.00a | 0.23 ± 0.00a | 0.10 ± 0.11a | 0.01 ± 0.01a | 0.01 ± 0.01a | 0.29 ± 0.08abc | 0.03 ± 0.05a | 0.00 ± 0.00b |
| | L | 0.01 ± 0.00a | 0.22 ± 0.03a | 0.06 ± 0.02a | 0.05 ± 0.00a | 0.00 ± 0.00a | 0.19 ± 0.09bc | 0.05 ± 0.02a | 0.08 ± 0.11ab |
| | A | 0.02 ± 0.02a | 0.18 ± 0.06a | 0.03 ± 0.00a | 0.03 ± 0.01a | 0.00 ± 0.01a | 0.23 ± 0.06bc | 0.08 ± 0.07a | 0.13 ± 0.04a |
| Jul. | C | 0.02 ± 0.00a | 0.08 ± 0.04b | 0.03 ± 0.00a | 0.05 ± 0.07a | 0.11 ± 0.16a | 0.22 ± 0.17bc | 0.11 ± 0.06a | 0.01 ± 0.01b |
| | L | 0.02 ± 0.01a | 0.18 ± 0.07ab | 0.04 ± 0.02a | 0.04 ± 0.03a | 0.12 ± 0.04a | 0.13 ± 0.02c | 0.11 ± 0.01a | 0.00 ± 0.00b |
| | A | 0.02 ± 0.00a | 0.14 ± 0.06ab | 0.04 ± 0.02a | 0.05 ± 0.00a | 0.12 ± 0.16a | 0.16 ± 0.01c | 0.06 ± 0.06a | 0.00 ± 0.00b |
| Aug. | C | 0.03 ± 0.03a | 0.20 ± 0.01a | 0.04 ± 0.04a | 0.01 ± 0.01a | 0.00 ± 0.00a | 0.24 ± 0.01abc | 0.15 ± 0.08a | 0.00 ± 0.00b |
| | L | 0.00 ± 0.00a | 0.18 ± 0.07ab | 0.06 ± 0.01a | 0.01 ± 0.01a | 0.00 ± 0.00a | 0.43 ± 0.14a | 0.15 ± 0.07a | 0.00 ± 0.00b |
| | A | 0.00 ± 0.00a | 0.20 ± 0.09a | 0.06 ± 0.01a | 0.02 ± 0.00a | 0.00 ± 0.00a | 0.37 ± 0.08ab | 0.15 ± 0.04a | 0.00 ± 0.00b |

[a] C, conventional; L, legume rotation; A, animal manure.

Table 5
Major fungal taxa by replicate. Values are mean ± standard deviation. Values within a column followed by different letters indicate significant differences ($P < 0.05$)

| Month | Treatment [a] | *Cladosporium* | *Fusarium* | *Gliocladium* | *Myrothecium* | *Penicillium* | *Trichoderma* | *Verticillium* |
|---|---|---|---|---|---|---|---|---|
| Jun. | C | 0.03 ± 0.00cd | 0.10 ± 0.00a | 0.10 ± 0.00ab | 0.10 ± 0.00b | 0.40 ± 0.00a | 0.12 ± 0.00ab | 0.03 ± 0.00d |
| | L | 0.04 ± 0.00bcd | 0.03 ± 0.01b | 0.06 ± 0.00bcd | 0.22 ± 0.10b | 0.38 ± 0.10a | 0.14 ± 0.05a | 0.06 ± 0.00cd |
| | A | 0.06 ± 0.03bc | 0.11 ± 0.09a | 0.10 ± 0.03abc | 0.22 ± 0.14b | 0.26 ± 0.01b | 0.13 ± 0.00ab | 0.01 ± 0.02d |
| Jul. | C | 0.06 ± 0.02bc | 0.06 ± 0.04ab | 0.14 ± 0.08a | 0.13 ± 0.06b | 0.17 ± 0.05c | 0.09 ± 0.02b | 0.11 ± 0.01abc |
| | L | 0.01 ± 0.00d | 0.02 ± 0.00b | 0.06 ± 0.03bcd | 0.63 ± 0.21a | 0.06 ± 0.04de | 0.02 ± 0.02c | 0.08 ± 0.07bcd |
| | A | 0.03 ± 0.01cd | 0.04 ± 0.03ab | 0.09 ± 0.02abcd | 0.13 ± 0.08b | 0.09 ± 0.02d | 0.05 ± 0.02c | 0.18 ± 0.05a |
| Aug. | C | 0.13 ± 0.03a | 0.04 ± 0.01ab | 0.02 ± 0.00cd | 0.54 ± 0.06a | 0.03 ± 0.01e | 0.03 ± 0.01c | 0.06 ± 0.04bcd |
| | L | 0.08 ± 0.02b | 0.03 ± 0.00b | 0.01 ± 0.01d | 0.55 ± 0.01a | 0.04 ± 0.00de | 0.02 ± 0.00c | 0.13 ± 0.04ab |
| | A | 0.04 ± 0.00bcd | 0.05 ± 0.02ab | 0.02 ± 0.00d | 0.66 ± 0.02a | 0.04 ± 0.00de | 0.02 ± 0.01c | 0.06 ± 0.01bcd |

[a] C, conventional; L, legume rotation; A, animal manure.

*lus* species. The pseudomonad population tended to decrease from June to July and then increase in August, as did the *Arthrobacter*, but most of these changes were not statistically significant. A cluster consisting of a variety of enteric bacteria, including *Klebsiella*, *Salmonella*, *Enterobacter*, *Rahnella*, *Hafnia*, and *Serratia*, increased throughout the season, but the increase was not statistically significant. Other genera observed included *Burkholderia* (mainly *Burkholderia cepacia*), *Flavobacterium* (*Flavobacterium indologenes*), *Curtobacterium*, *Staphylococcus*, *Micrococcus*, *Clavibacter*, *Rathayibacter*, *Cellulomonas*, *Rhodococcus*, *Yersinia*, *Morganella*, *Corynebacterium*, and *Sphingobacterium*.

A total of 40 fungal genera were identified. Major fungal taxa are presented in Table 5. Penicillia dominated in June and *Myrothecium* in July and August. Myrothecia were remarkably dominant in August, which explains the lower diversity in August. In June, significant treatment effects were observed for fusaria and penicillia, while in July significant treatment effects were found for myrothecia and penicil-

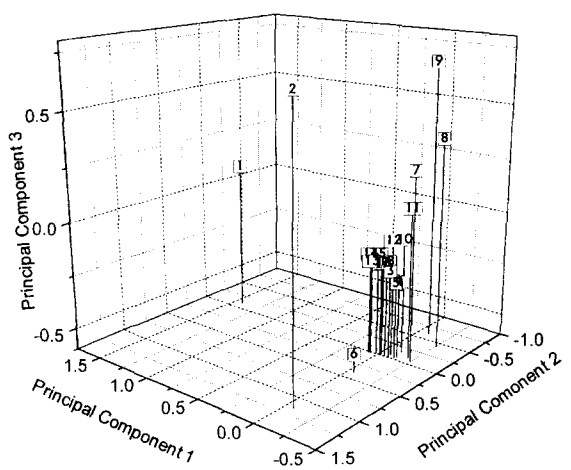


Fig. 1. Principal component analysis of bacterial taxa distribution. 1–6, June; 7–12, July; 13–18, August; 1, 7, 13, conventional replicate 5; 2, 8, 14, legume replicate 5; 3, 9, 15, animal manure replicate 5; 4, 10, 16, legume replicate 4; 5, 11, 17, animal manure replicate 4; 6, 12, 18, conventional replicate 4.

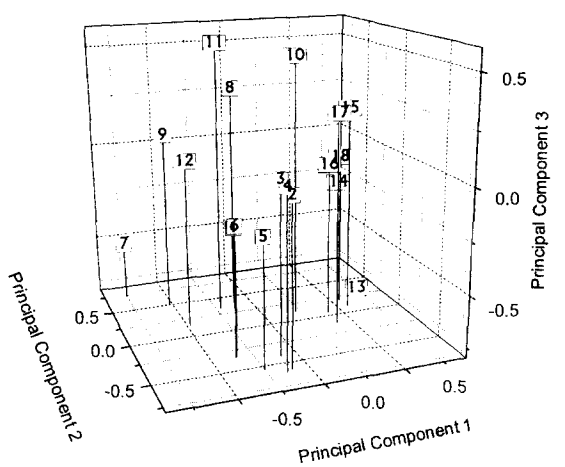


Fig. 2. Principal component analysis of fungal taxa distribution. 1–6, June; 7–12, July; 13–18, August; 1, 7, 13, conventional replicate 5; 2, 8, 14, legume replicate 5; 3, 9, 15, animal manure replicate 5; 4, 10, 16, legume replicate 4; 5, 11, 17, animal manure replicate 4; 6, 12, 18, conventional replicate 4.

lia. Other taxa observed included *Absidia*, *Alternaria*, *Cephalosporium*, *Circinella*, *Hughesiella*, *Humicola*, *Rhizopus*, *Stachybrotrys*, and *Stemphyllium*.

Principal component analysis of the relative abundance of the bacterial and fungal taxa was performed (Figs. 1 and 2). The replicates do not cluster by treatment, but do show some tendency to cluster by month. We conclude, again, that the seasonal effect was greater than any treatment effect.

## 4. Discussion

In this study diversity and evenness were measured using isolates. This method has the disadvantage of relying on culturable organisms, which may represent a small fraction of the total population. Furthermore, it is impossible to know whether fungal isolates are due to active mycelia or spore germination. However, it does have the advantage of allowing us to identify the microorganisms and examine their biochemistry (Palleroni, 1994).

We used two different diversity and evenness indices to make sure our results were not an artifact of the index used. Shannon's index is based on a mathematical theory of information, and can be regarded as an uncertainty measure (Pielou, 1969). If we choose an individual at random from a population with higher diversity (a greater number of taxa with more even distribution of individuals among the taxa) we will be more uncertain as to which taxon that individual belongs to. McIntosh's diversity index can be viewed geometrically. The population is considered to be a point in $S$-dimensional space, where $S$ is the number of taxa, and the coordinates of the point will be $N_1 \ldots N_S$ (McIntosh, 1967; Pielou, 1969). The Euclidean distance of this point from the origin is $\sqrt{\sum_i N_i^2}$. As species richness and evenness increase, the point representing the population will move closer to the origin, so the Euclidean distance will decrease.

The three treatments, conventional, legume, and animal manure, had very little effect on microbial diversity, evenness, or dominance of specific taxa. The conventional system used a corn–soybean rotation, and results might be different for a monoculture system with continuous corn. Only rhizosphere organisms were studied, and it may be that treatment effects would be larger for microorganisms in the bulk soil. A study on isolates from rhizosphere and root-free soil demonstrated larger seasonal effects on rhizosphere than bulk soil isolates and, as in this paper, the seasonal effects were greater than treatment effects (Hassink et al., 1991). It may also be that treatment effects would have been observed if a different method, such as fatty acid or PCR-RFLP analysis of rhizosphere soil, had been performed. However, phospholipid analysis of bulk soil taken from the Farming Systems Trial at Rodale in November 1991 did not show any statistically significant treatment effects, because differences between replicates were as large as those between treatments (Wander et al., 1995), which is consistent with our results.

The Farming Systems Trial was studied during the second and fifth years after conversion from conventional management (Doran et al., 1987). Fungal and bacterial populations were greatest in fields planted in red clover during the second year, but the management system had no effect on population when similar crops (corn or soybean) were compared. Plots planted in hairy vetch in the fifth year had greater microbial biomass than other plots.

A number of studies have found treatment effects on microbial diversity or on relative population size of specific taxonomic groups (Liljeroth et al., 1990; Boehm et al., 1993; Frostegård et al., 1993; Kirchner et al., 1993; Kennedy and Smith, 1994; Workneh and van Bruggen, 1994; Zelles et al., 1994). However, each study used different methods and studied different systems, so it is impossible to generalize or to directly compare these studies to ours. We are currently identifying actinomycetes that were isolated at the same time as the bacteria and fungi reported on in this study. We are also performing fatty acid analysis on root zone soil that was collected and frozen during this study. Finally, selected bacterial isolates from the various taxa are being analyzed for protease, cellulase, and inorganic phosphatase activity. These studies may allow us to more directly compare our results to other studies reported in the literature, and will also provide a direct comparison between phenotypic analysis of isolates and fatty acid analysis of soil.

## Acknowledgements

We thank Stan Tesch, Heidi Hartman, Joshua Olsen, Betsy Lyman, and Leeanne Teerlinck for technical assistance. We thank former and current members of the Rodale Institute Research Center for their work in maintaining the Farming Systems Trial, particularly Rhonda Janke, Steve Peters, and Laurie Drinkwater.

## References

Boehm, M.J., Madden, L.V. and Hoitink, H.A.J., 1993. Effect of organic matter decomposition level on bacterial species diversity and composition in relationship to *Pythium* damping-off severity. Appl. Environ. Microbiol., 59: 4171–4179.

Buyer, J., 1995. A soil and rhizosphere microorganism isolation and enumeration medium that inhibits *Bacillus mycoides*. Appl. Environ. Microbiol., 61: 1839–1842.

Cook, R.J. and Baker, K.F., 1983. The Nature and Practice of Biological Control of Plant Pathogens. American Phytopathological Society, St. Paul, MN.

Doran, J.W., Fraser, D.G., Culik, M.N. and Liebhardt, W.C., 1987. Influence of alternative and conventional agricultural management on soil microbial processes and nitrogen availability. Am. J. Altern. Agric., 2: 99–106.

Frostegård, A., Bååth, E. and Tunlid, A., 1993. Shifts in the structure of soil microbial communities in limed forests as revealed by phospholipid fatty acid analysis. Soil Biol. Biochem., 25: 723–730.

Gilman, J.C., 1957. A Manual of Soil Fungi, 2nd edn. The Collegiate Press, Ames, IA, 450 pp.

Hassink, J., Oude Voshaar, J.H., Nijhuis, E.H. and van Veen, J.A., 1991. Dynamics of the microbial populations of a reclaimed-polder soil under a conventional and a reduced-input farming system. Soil Biol. Biochem., 23: 515–524.

Kaufman, D.D., Williams, L.E. and Sumner, C.B., 1963. Effect of plating medium and incubation temperature on growth of fungi in soil-dilution plates. Can. J. Microbiol., 9: 741–751.

Kennedy, A.C. and Smith, K.L., 1994. Soil microbial diversity and ecosystem functioning. In: G.P. Robertson and H. Collins (Editors), The Functional Significance and Regulation of Soil Biodiversity. Kluwer, the Netherlands.

Kirchner, M.J., Wollum, II, A.G. and King, L.D., 1993. Soil microbial populations and activities in reduced chemical input agroecosystems. Soil Sci. Soc. Am. J., 57: 1289–1295.

Lee, K.E., 1994. The biodiversity of soil organisms. Appl. Soil Ecol., 1: 251–254.

Liljeroth, E., Schelling, G.C. and van Veen, J.A., 1990. Influence of different application rates of nitrogen to soil on rhizosphere bacteria. Neth. J. Agric. Sci., 38: 255–264.

Magurran, A., 1988. Ecological Diversity and its Measurement. Croom Helm, London, 179 pp.

McIntosh, R.P., 1967. An index of diversity and the relation of certain concepts to diversity. Ecology, 48: 392–404.

O'Donnell, A.G., Goodfellow, M. and Hawksworth, D.L., 1994. Theoretical and practical aspects of the quantification of biodiversity among microorganisms. Phil. Trans. R. Soc. London, Ser. B, 345: 65–73.

Palleroni, N.J., 1994. Some reflections on bacterial diversity. ASM News, 60: 537–540.

Parr, J., Papendick, R., Hornick, S. and Meyer, R., 1992. Soil quality: Attributes and relationship to alternative and sustainable agriculture. Am. J. Altern. Agric., 7: 5–11.

Pielou, E.C., 1969. An Introduction to Mathematical Ecology. John Wiley, New York, 286 pp.

Raper, K.B. and Thom, C., 1949. Manual of the Penicilia. Williams and Wilkins, Baltimore, MD, 875 pp.

Thom, C. and Raper, K.B., 1945. Manual of the Aspergilli. Williams and Wilkins, Baltimore, MD, 373 pp.

Visser, S. and Parkinson, D., 1992. Soil biological criteria as indicators of soil quality: Soil microorganisms. Am. J. Altern. Agric., 7: 33–37.

Wander, M.M., Hedrick, D.S., Kaufman, D., Traina, S.J., Stinner, B.R., Kehrmeyer, S.R. and White, D.C., 1995. The functional significance of the microbial biomass in organic and conventionally managed soils. Plant Soil, 170: 87–97.

Workneh, F. and van Bruggen, A.H.C., 1994. Microbial density, composition, and diversity in organically and conventionally managed rhizosphere soil in relation to suppression of corky root of tomatoes. Appl. Soil Ecol., 1: 219–230.

Zelles, L., Bai, Q.Y., Ma, R.X., Rackwitz, R., Winter, K. and Beese, F., 1994. Microbial biomass, metabolic activity and nutritional status determined from fatty acid patterns and poly-hydroxybutyrate in agriculturally managed soils. Soil Biol. Biochem., 26: 439–446.